

A NEW CLASS OF HALOGENATED LACTONES FROM THE RED ALGA
DELISEA FIMBRIATA (BONNEMAIISONIACEAE)

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Several species within genera of the family of red algae Bonnemaisoniaceae have yielded halogenated secondary metabolites. Halogenated heptan-2-ones were isolated from *Bonnemaisonia hamifera*¹ and two independent studies of geographically separate collections of *Asparagopsis taxiformis*^{2,3} yielded haloacetones or haloforms respectively as major isolates whilst halo-butenones were detected in both studies. We have found haloacetones, including the previously unreported 1,1-dibromo-3-iodoacetone, together with bromoform in a freeze-dried sample of *A.armata* from Australian waters⁴ but could detect no halobutenones; also 1,1,2-tribromooctenones of general formula (1) were isolated from the dichloromethane extract of a Tasmanian collection of *Ptilonia australasica*⁵.

Our interest in *Delisea fimbriata* (Bonnemaisoniaceae)⁶ was stimulated by the significant *in vitro* antimicrobial and antifungal activity of the dichloromethane extract of the freeze-dried alga collected in many locations near Sydney. Individual collections, either extracted immediately (wet) or after freeze-drying of freshly collected material, yielded 5% (dry weight) of a complex mixture of dichloromethane soluble material which was resolved by silica gel chromatography into three major fractions which demonstrated a common u.v. chromophore at ca. λ_{\max} 290 nm.

G.c./m.s. data on these fractions revealed that each was a complex mixture but separate components of each fraction could be rationalised by the general formula $C_9H_9O_2BrRXY$ with X,Y either hydrogen or halogen (Br,Cl,I). For the least polar fraction where R=H (4a-c) (14% of the crude extract), we propose the generic name 'fimbrolide'. The acetoxyfimbrolides (R=OAc) (2a-g) represented 36% of the dichloromethane soluble material. The most polar fraction, the hydroxyfimbrolides (R=OH) (3a-g) represented 14% of the mixture.

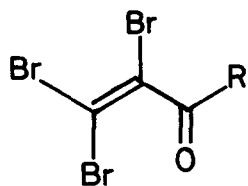
The general structures (2), (3) and (4) for the whole series of halogenated metabolites was deduced from the following evidence. The ¹H n.m.r. spectrum (CDCl₃)⁷ of the mixture of acetoxyfimbrolides (2) showed resonances

corresponding to a $\text{CH}_3\text{CH}_2\text{CH}_2\text{CH}(\text{OAc})-\overset{1}{\text{C}}=$ group [$\delta 0.94$ (3H, t, $J=7\text{Hz}$); 1.34 (2H, m); 1.86 (2H, m); 2.09 (3H, s) and 5.52 (1H, t)] together with two sharp singlets at $\delta 6.70$ and 6.38 (1H combined, ratio 1:3) suggestive of a mixture of E and Z isomers. The mixture could be resolved into the E and Z isomers (2a) and (2b) (contaminated with the other minor components) by HPLC. The i.r. spectrum showed the presence of two carbonyl groups at 1780 cm^{-1} (γ -lactone) and 1740 cm^{-1} (acetate) and the u.v. spectrum (MeOH) $\lambda_{\text{max}}^{18} 293\text{nm}$ ($E_{1\text{cm}}^{18} = 220$) demanded that the γ -lactone function was doubly conjugated. Zinc-acetic acid reduction of the acetate mixture produced a single keto-acid in high yield. The ^1H n.m.r. of this acid [$\delta 7.09$ (1H, t, $J=8\text{Hz}$); 3.39 (2H, s); 2.16 (3H, s); 2.10 (2H, m); 1.48 (2H, m) and 0.92 (3H, bt)] demanded either structure (5) or (6). Reduction of this acid with sodium borohydride gave the product (7) [^1H n.m.r.: $\delta 7.02$ (1H, t); 5.86 (2H, bs; D_2O exchangeable); 3.92 (1H, m); 2.42 (2H, d, $J=6\text{Hz}$); 2.14 (2H, m); 1.42 (2H, m); 1.22 (3H, d, $J=6\text{Hz}$) and 0.95 (3H, bt)] which could only be formed from (5). Thus the major proportion of the mixture of acetoxyfimbrolides was (2a) and (2b).

The presence of (2a,b) (m/e 370, 368, 366; $\text{C}_{11}\text{H}_{12}\text{O}_4\text{Br}_2$), (2c,d) (m/e 416, 414; $\text{C}_{11}\text{H}_{12}\text{O}_4\text{BrI}$), (2e,f) (m/e 326, 324, 322; $\text{C}_{11}\text{H}_{12}\text{O}_4\text{BrCl}$) and (2g) (m/e 450, 448, 446, 444; $\text{C}_{11}\text{H}_{11}\text{O}_4\text{Br}_3$) were detected by g.c./m.s. of the gross mixture of acetoxyfimbrolides and high resolution m.s. measurements confirmed the elemental compositions. The position of chlorine or iodine in these minor products at C5 was established by addition of diethylamine to an ethereal solution of the gross mixture of acetoxyfimbrolides (2a-g). This afforded a mixture of the enamines (8) and (9). Mass spectral analysis of the enamine mixture showed that it consisted entirely of monobromo-compounds and no chloro- or iodo-analogues were detected. The u.v. spectrum of this mixture $\lambda_{\text{max}}^{265} 265, 285$ (s), $373, 388\text{nm}$ ($\log \epsilon 3.70, 3.66, 4.10, 4.13$) supported the proposed structure thereby establishing that the halogen substituent at C3 was bromine in all compounds. The ^1H n.m.r. spectrum (CDCl_3) of the mixture of (8) and (9) showed two low field singlets at $\delta 6.28$ and 6.56 (ratio 9:1; 1H combined). In the most stable isomer (8) the proton at C5 was at higher field in the ^1H n.m.r. spectrum than in (9) thus, by analogy, it was surmised that Z-acetoxyfimbrolide (2b) was the major compound in the mixture of acetoxyfimbrolides.

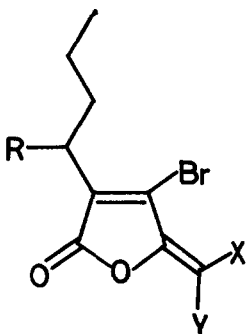
The E and Z hydroxyfimbrolides (3a) and (3b) could not be separated by silica gel chromatography. The ^1H n.m.r. spectrum (CDCl_3) showed resonances at $\delta 6.70, 6.36$ (1H combined, both s, ratio 1:2), 4.60 (1H, t), 2.96 (1H, bs; D_2O exchangeable), 1.78 (2H, m), 1.40 (2H, m), 0.94 (3H, t). G.c./m.s. showed (3a,b) (m/e 328, 326, 324; $\text{C}_9\text{H}_{10}\text{O}_3\text{Br}_2$); (3c,d) (m/e 374, 372; $\text{C}_9\text{H}_{10}\text{O}_3\text{BrI}$); (3e,f) (m/e 284, 282, 280; $\text{C}_9\text{H}_{10}\text{O}_3\text{BrCl}$) and (3g) (m/e 408, 406, 404, 402; $\text{C}_9\text{H}_9\text{O}_3\text{Br}_3$).

The least polar fraction contained the fimbrolides (4), the



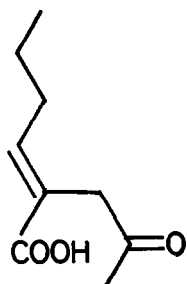
- R = CH₂Br
- R = CH⁺CHCBr₂CH₂CH₃
- R = CHBr CH₂CH₂CH₂CH₃
- R = CBr₂ CH₂CH₂CH₂CH₃

(1)

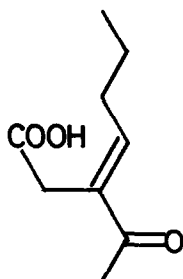


	X	Y	% of dry alga *	
(2) R = OAc	(a)	Br H	0.32	
	(b)	H Br	1.01	
	(c)	I H)	0.15	
	(d)	H I)		
	(e)	Cl H)		
	(3) R = OH	(f)	H Cl)	0.11
		(g)	Br Br	0.10
(a)		Br H	0.17	
(4) R = H	(b)	H Br	0.34	
	(c)	I H)	0.04	
	(d)	H I)		
	(e)	Cl H)		
	(f)	H Cl)	0.04	
(g)	Br Br	0.01		
(4) R = H	(a)	Br H	0.10	
	(b)	H Br	0.17	
	(c)	Br Br	0.38	

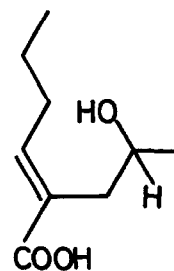
* Estimated by GC (FID detector)



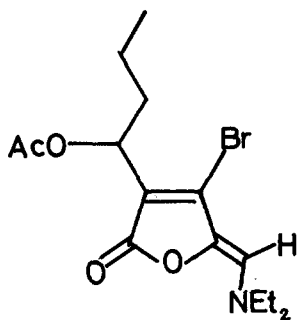
(5)



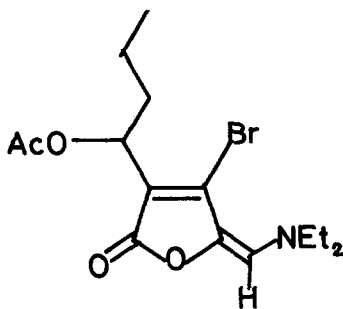
(6)



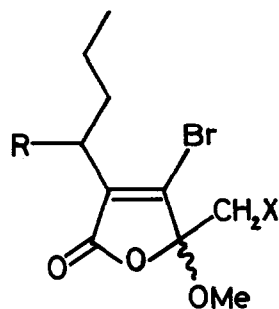
(7)



(8)



(9)



(10)

^1H n.m.r. spectrum (CCl_4) of which showed resonances at δ 6.56 and 6.22 (0.4H comb., both s, ratio 1:2), 2.40(2H, t), 1.40(4H, m), 0.92(3H, t). G.c./m.s. showed (4a,b) (m/e 312, 310, 308; $\text{C}_9\text{H}_{10}\text{O}_2\text{Br}_2$) and (4c) (m/e 392, 390, 388, 386; $\text{C}_9\text{H}_9\text{O}_2\text{Br}_3$).

Brief treatment of a methanolic solution of the acetoxyfimbrolides (2a-g) with methanolic KOH followed by immediate acidification with HCl produced a mixture of methanol addition products (10, R=OAc). Compound (10, R=OH) was also formed from (3) under the same conditions.

Storage of freeze-dried samples of *D.fimbriata* at room temperature for 6 months resulted in an appreciable change in the composition of the extract. Trace amounts of (3) were obtained and the yield of (2) and (4) were 0.54% and 0.22% of dry weight of the organism respectively. Compound (2a) was absent from the mixture of acetoxyfimbrolides which consisted mainly of (2b) and (2d) (ratio 5:2). This would suggest that (2a,b) decompose more readily than the iodo-analogue (2d) in the freeze dried organism. This result is similar to that obtained by Sims *et al*⁸ from air dried samples of *D.fimbriata* collected from Antarctic waters.

The biosynthesis of these compounds is rather uncertain but they appear to be derived either by a mixed acetate pathway or from degradation of a phloroglucinol derivative. This will be investigated in the near future.

References:

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- 6) Levring and Tore, *Arkiv. for Botanik. Ser 2*, 11, No.6, 457-530 (1953) gives *D.fimbriata* priority in name over the alternative name *D.pulchra*.
- 7) The ^1H n.m.r. spectrum of the Z-acetoxyfimbrolides varied appreciably with solvent. For the values in CCl_4 see following paper⁸.
- 8) J.A.Pettus, Jr., R.M.Wing and J.J.Sims, accompanying paper.